# ALASKA
# HARBOR SEAL RESEARCH PLAN

**2000**

**National Marine Fisheries Service**
Alaska Region, Protected Resources Division
Alaska Fisheries Science Center, National Marine Mammal Laboratory
Southwest Fisheries Science Center

**Alaska Department of Fish and Game**
Division of Wildlife Conservation

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**Introduction** . . . . . 1

**Management Needs** . . . . . 2

**Goals of the Harbor Seal Research Plan** . . . . . 2

**A. ABUNDANCE AND TREND ESTIMATION** . . . . . 3
1. Aerial surveys to estimate population abundance . . . . . 4
2. Correction factor study . . . . . 5
3. Aerial surveys to estimate population trends . . . . . 6
**4. Covariate analysis** . . . . . 7
5. Haulout patterns and movement . . . . . 8
6. Glacial survey methodology . . . . . 9
7. Index site counts . . . . . 10
8. Evaluation and review of survey experimental design . . . . . 11

**B. STOCK IDENTIFICATION** . . . . . 12
1. mtDNA variation among and within stock strata defined ***a priori*** . . . . . 13
2. *A posteriori* determination of mtDNA stock boundaries . . . . . 15
3. Stock identification - microsatellites . . . . . 16
4. Estimates of genetic diversity as indicators of population fitness . . . . . 18
5. Alternative approaches to sample acquisition . . . . . 19
6. Harbor seal mating systems and dispersal patterns . . . . . 20

**C. HARBOR SEAL HABITAT** . . . . . 21
1. Characterize harbor seal habitat . . . . . 21

**D. HEALTH AND CONDITION** . . . . . 22
1. Contaminant analysis . . . . . 22
2. Disease/Pathology . . . . . 24
3. Health Index . . . . . 25

**E. FOOD HABITS** . . . . . 26
1. Food item analysis . . . . . 26
2. Fatty acid analysis . . . . . 27

**F. LIFE HISTORY/GENERAL BIOLOGY** . . . . . 29
1. Pupping and molting phenology . . . . . 29
2. Pupping colony(ies) in SE Alaska . . . . . 30

**G. VITAL RATES** . . . . . 31

1. Female reproductive biology . . . . . 31
2. Estimating age, age of maturation, and indices of growth from teeth . . . . . 32
3. Individual identification . . . . . 33

**H. HUMAN INTERACTIONS** . . . . . 34
1. Disturbance at haulouts . . . . . 34
2. Harvest monitoring and mortality estimation . . . . . 35
3. Incidental take by commercial fisheries . . . . . 36

**Acknowledgements** . . . . . 38

**Literature Cited** . . . . . 39

**Appendix 1** . . . . . 46

**Introduction**

Harbor seals, *Phoca vitulina,* are one of the most widely distributed pinnipeds in coastal Alaska, ranging from the southern part of the state at Dixon Entrance, west to the Aleutian Islands and into the southern Bering Sea. Harbor seals represent a significant marine resource to a range of users: subsistence harvesters, visitors, local Alaska residents, and others. Once considered abundant throughout their range, counts of harbor seals have declined in some areas of the state over the last 20 years.

The decline in numbers is most apparent in the Kodiak Archipelago of the Gulf of Alaska, as well as in Prince William Sound (PWS). Tugidak Island, in the Kodiak Archipelago, has historically been the focus of much of the research directed at harbor seals, and has yielded a large amount of information on this pinniped in Alaska. The number of harbor seals at Tugidak Island declined by approximately 90% from the mid-1970s through the early 1990s (Pitcher 1990, Small *et al*. 1998). Counts of harbor seals in Prince William Sound declined by 63% between 1984 and 1997 (Frost et al. 1998). Other pinniped species in Alaska, notably Steller sea lions and fur seals, have also experienced a decline in numbers over roughly the same time period. Concern for the status of harbor seals on a local and statewide basis highlights the need for continued and expanded research on this species in Alaska.

Research on Alaska harbor seals that is funded through the National Oceanic and Atmospheric Administration's National Marine Fisheries Service (NMFS) is carried out principally by three groups: NMFS, Alaska Fisheries Science Center's National Marine Mammal Laboratory (NMML); Alaska Department of Fish and Game, Division of Wildlife Conservation (ADF&G); and NMFS, Southwest Fisheries Science Center (SWFSC). Research efforts by the above-mentioned institutions support management needs directed at conserving healthy harbor seal populations in Alaska. Within NMFS, the management responsibility for harbor seals in Alaska lies with the Alaska Region, Protected Resources Division.

The combined research efforts by NMFS and ADF&G focus on statewide harbor seal population abundance estimation, stock identification, trends in abundance, general biology and life history, and human interactions. Additional research on Alaska harbor seals is also being conducted by independent researchers, University researchers, and by the National Park Service in Glacier Bay National Park and Preserve.

Native Alaskans traditionally hunt harbor seals for subsistence food and handicrafts and have an accumulation of traditional knowledge associated with this species. The Alaska Native Harbor Seal Commission (ANHSC) represents Native interests on matters associated with harbor seals. On April 29, 1999, NMFS and the ANHSC signed an agreement as partners in the co- management of the subsistence uses of harbor seals in Alaska (NMFS/ANHSC 1999).

While there are a number of entities involved in both research and management issues concerning

harbor seals in Alaska, the content of this Alaska Harbor Seal Research Plan is restricted to research funded by NMFS and conducted by NMFS and ADF&G.

**Management Needs**

As mandated by the Marine Mammal Protection Act of 1972 as Amended (MMPA), NMFS is required to maintain the health and stability of marine ecosystems. Consistent with this major goal, three explicit objectives of the MMPA are to 1) maintain stocks at their optimum sustainable population (OSP) levels and as functioning elements of their ecosystems, 2) restore depleted stocks to OSP levels and 3) reduce incidental mortality and serious injury (from commercial fisheries) to "insignificant levels approaching a zero mortality and serious injury rate" (MMPA 1995, Barlow et. al 1995).

Therefore, the fundamental objective of management is to prevent "depletion" of a species or population stock or to restore a species or population stock to its OSP. A species or population is said to be depleted when the Secretary of Commerce "determines that a species or population stock is below its optimum sustainable population [OSP]." OSP is "the number of animals which will result in the maximum productivity of the population or the species, keeping in mind the carrying capacity of the habitat and the health of the ecosystem of which they form a constituent element" (MMPA 1995).

To meet the overall management objectives as mandated by the MMPA, specific information must be available to managers. Management must be able to identify and describe a given group of animals (i.e., a stock or population of harbor seals); describe the status of this unit, including minimum population estimates and the trend in numbers; estimate human-induced mortality levels, including those resulting from commercial fisheries and subsistence removals; as well as have knowledge of the species' biology and ecology to determine how the species may be affected and by what factors. The principal categories of research presented in this plan are designed to provide information to meet the management objectives described above. Using this information, management is tasked with designing appropriate measures that will mediate negative impacts to harbor seals.

In addition, NMFS has recently entered into a new management partnership with the ANHSC to co-manage the subsistence use of harbor seals in Alaska. The agreement between NMFS and the ANHSC includes, as a principal objective, to provide for the maintenance of harbor seal population levels that will allow for long-term sustainable harvests. Information collected by the various research projects will also contribute towards ensuring that this management objective can be met.

**Goals of the Harbor Seal Research Plan**

Prior to the development of this document no consolidated research plan for all NMFS-funded harbor seal research, which is carried out by several institutions, existed. The current document is intended to consolidate all of these research efforts. A consolidated plan will provide the opportunity for a more effective evaluation of existing Alaska harbor seal research. It will provide a platform for modifications

to be made to this research based on priorities that satisfy management objectives. The timescale is intended to be a five-year outlook; although the document will be revised annually. A five-year timescale was chosen to fit with budgeting cycles within the agency and to accommodate the full cycle of aerial surveys for the complete range of harbor seals in Alaska.

The principal objectives of this Research Plan are the following:
1) consolidate the research plans of currently-funded projects into a single coordinated effort;
2) describe additional research projects that are currently unfunded, but for which funds are being sought;
3) increase the dialogue, coordination, and collaboration among NMFS-funded harbor seal researchers and NMFS managers through the process of annually reviewing, evaluating and updating the Research Plan; and
4) ensure that research on Alaska harbor seals satisfies management objectives.

The Research Plan will also serve a useful role in the co-management process by drawing together the current and proposed descriptions of harbor seal research projects funded by NMFS. In doing so, the plan should enhance communication between NMFS and the ANHSC by describing NMFS-funded research plans and priorities. Similarly, the plan is expected to provide helpful information to the Alaska Scientific Review Group about NMFS-funded harbor seal research.

The Research Plan has been organized according to broad research categories with individual projects outlined in greater detail. Each task includes a research category overview, project objectives, justification, methods, product, five-year project status and project lead. The scope of research projects includes both short- and long-term tasks, depending on the nature of the investigation. This Research Plan will be evaluated and revised annually as research and management objectives are met, needs evolve and funding commitments change.

## Research Categories

## A. ABUNDANCE AND TREND ESTIMATION

### Overview

This research category comprises 8 tasks that relate to estimation of abundance and trends in population size of Alaska harbor seals. Because the interdependence of the tasks is complex and important to the justification for the research, a brief overview seems appropriate. Two of the tasks, numbered 1 and 3, describe aerial surveys to count harbor seals on their haulout grounds. Task 1 describes aerial surveys conducted by NMML to estimate statewide abundance of harbor seals on about a 5-year cycle. Task 3 describes aerial surveys conducted by ADF&G to estimate trends of

harbor seal populations in selected regions of the state. These tasks differ in their objectives and geographic scope, but are subject to the same factors that introduce variability in the proportion of the seal population that is ashore to be observed and counted during the surveys. Therefore, both survey tasks are subject to the same constraints and difficulties of designing the surveys to either minimize variability from factors such as tides, weather conditions, time of day, and date (in the seals' seasonal life history), or to adjust for the state of those factors during the surveys. Task 4, Covariate Analysis, is an investigation of methods for the latter approach of adjusting for factors that affect the availability of seals to be counted. This approach is being tested on data from both the ADF&G and the NMML aerial surveys. Even when counts are adjusted for covariates, estimating the total population size requires estimating the proportion of the population that was not included in the survey because some seals were unavailable to be counted. Task 2 is concerned with estimating a correction factor for the proportion missed and determining the effects of different haulout substrates (e.g., rock, sand, ice) on the proportion missed. With the development of techniques for covariate adjustment, special care may have to be taken to ensure that the correction factors can be estimated in a way that is compatible with covariate-adjusted counts. Several of the remaining tasks also provide support for modeling covariate relationships and estimating correction factors, especially Task 5, which documents the haulout patterns of harbor seals, and task 7, which involves frequent and regular counting of seals hauled out at Tugidak Island. Task 6 is a special effort to address practical difficulties of surveying harbor seals that use glacial ice for hauling out. Finally, task 8 is a study using simulation models to investigate the efficacy and robustness of the design for harbor seal surveys, primarily in the *Exxon Valdez* oil spill area (i.e. Prince William Sound southwest to Tugidak Island), but with applications statewide. This study will provide important guidance for the overall approach by NMFS and ADF&G, described here, for gathering critical information on the abundance and trends of harbor seals in Alaska.

**1. Aerial surveys to estimate population abundance**

**Objective:** Estimate statewide distribution and abundance of harbor seals in Alaska.

**Justification:** The 1994 amendments to the Marine Mammal Protection Act (Section 117) require that NMFS produce a Stock Assessment Report (SAR) on the status of each species under its jurisdiction. Certain key population parameters are required to describe the status of the stock, including population size. Minimum population estimates are also needed for the calculation of a Potential Biological Removal (PBR) level, also required in the SARs. Since 1991 the NMFS National Marine Mammal Laboratory, in cooperation with ADF&G, has conducted yearly censuses of harbor seals in Alaska to provide baseline population information and for purposes of calculating a minimum population estimate for the SARs.

**Methods:** For purposes of conducting abundance surveys, the state of Alaska has been divided into 5 survey regions, corresponding approximately to the stock boundaries described in the SARs: (1) north side of the Alaska Peninsula and Bristol Bay; (2) Aleutian Islands; (3) Gulf of Alaska; (4) northern Southeast Alaska; and (5) southern Southeast Alaska. Each subdivision is surveyed on a rotating schedule. Logistics and resources preclude the completion of a more comprehensive survey on a more

frequent basis. Two complete abundance estimates are, therefore, produced every decade. It should be noted that when zones already surveyed as part of the aerial survey trend routes are present in the survey area, these zones are not double-counted (i.e. the counts from the trend routes are incorporated in the abundance estimates).

Fixed-wing aircraft are used to photograph harbor seals hauled out on land or on ice during the molt season in August. Aerial surveys are flown between 100 and 300m altitude at 90 knots, within 2 hours on either side of low tide. Covariate analyses consistently show that date, time relative to mid-day, and time of day strongly influence counts, whereas tide height has less influence. The entire coastline is surveyed several times initially to identify haulout sites, then observers fly from site to site including all previously known haulouts. Four to six repetitive counts on separate days are planned for each major haulout site within each study area over the 2 week survey period.

Harbor seals are photographed with 35 mm cameras equipped with a 70-210 mm lens using ASA 200 or 400 color slide film. Transparencies are projected onto a white background and the number of seals counted. Two counters will score the number of seals on the photographs for each area for each survey day and the arithmetic mean will be calculated for each site. The largest arithmetic mean obtained for each area is used as the minimum population estimate. For small groups of seals hauled out, visual estimates are obtained.

**Product:** The total abundance of harbor seals statewide and for each of the regions will be estimated on about a 5-year cycle. These estimates will comprise minimum population sizes in the form of the actual numbers of seals counted on aerial surveys, counts adjusted for covariates, and estimates of abundance obtained by correcting for the proportion of the population unavailable to be counted during the aerial surveys.

**Five-year project status:** Area-specific abundance estimates will be produced according to the following schedule (the rotation schedule will be repeated):

| | |
|---|---|
| Y2000 | north side of the Alaska Peninsula and Bristol Bay |
| Y2001 | Gulf of Alaska |
| Y2002 | northern Southeast Alaska |
| Y2003 | southern Southeast Alaska |
| Y2004 | Aleutian Islands |

Funded FY00.

**Project lead:** NMML

**2. Correction factor study**

**Objective:** Estimate the proportion of seals not counted during aerial abundance surveys for calculation of a "correction factor" which is applied to counts collected during those surveys to

determine population estimates.

**Justification:** Harbor seals are censused from aircraft by photographing the seals hauled out on land or ice during the molt period (August/September). These surveys miss an unknown number of animals that are at sea during the survey period. A specific "correction factor" has been developed for rocky, sandy, and ice substrates and is applied to census data to obtain a minimum population estimate for publication in the Stock Assessment Reports (SARs) and for use in the calculation of a Potential Biological Removal (PBR) for a given population. Previously, one correction factor was applied to all census estimates irrespective of substrate. The results of the correction factor estimation studies show that for seals hauling out on rocky substrates, the estimated number of seals from aerial abundance surveys should be multiplied by 1.74 to provide a better estimate of the true number of animals present (research completed in 1994, SE Alaska). For seals hauling out on sandy substrates, aerial counts should be multiplied by 1.90 (research conducted in 1995 & 1996, Prince William Sound). For seals hauling out on ice (glacial) substrates, aerial counts should be multiplied by 1.92 (research conducted in 1997 & 1998, Kenai Peninsula glaciers). A group of scientific experts and the AKSRG recommended that correction factor studies be expanded to produce estimates not only for the various substrate types, but also for each survey region. Therefore, future correction factor studies will occur in the same region as our abundance surveys to better understand survey timing and those factors (covariates) affecting seal haulout behavior.

**Methods:** Several weeks prior to annual assessment surveys (August), up to 50 seals will be captured and outfitted with flipper (rear) mounted VHF transmitters. During the aerial assessment surveys, approximately 8-10 replicate flights will be flown and the number, identification, and location of tagged seals hauled out during each survey will be determined. These data are used in a mark-recapture framework to estimate the correction factor for seals missed because they were not hauled out during the survey. Counts from the aerial assessment surveys will be multiplied by the correction factor, yielding a more accurate estimate of the number of seals in an area.

**Product:** This study will provide area-, year-, and substrate-specific factors to correct harbor seal counts for seals missed during aerial surveys.

**Five-year project status:** Current plans call for conducting this project annually using the same schedule as the aerial assessment surveys for abundance. In August 2000, this work will be conducted in Bristol Bay. Funded FY00

**Project lead:** NMML

**3. Aerial surveys to estimate population trends**

**Objective:** Estimate population trends for harbor seals in the areas of Ketchikan, Sitka, Prince William Sound (PWS), the Kodiak Archipelago and Bristol Bay.

**Justification:** Changes in harbor seal numbers over time have been monitored through aerial trend surveys that estimate the number of seals (i.e., counts) at designated sites within selected survey routes distributed throughout the state (Frost *et al*. 1999, Small *et al*. 1999). Trend surveys are aimed at determining whether harbor seal numbers are increasing, stable, or decreasing in different parts of their range in Alaska. The locations of trend routes currently monitored were selected to be generally representative of harbor seals throughout the state. This trend information is fundamental to an understanding of population status (Gerber *et al*. 1999) and to the implementation of the PBR approach to management under the MMPA. Continued monitoring of these routes will allow researchers and managers to follow population trends and make adjustments to research objectives, as well as to implement management measures that would assist in recovery of depressed populations and to monitor the effectiveness of management measures.

**Methods:** Standardized aerial survey techniques will be used to count animals at all haulout sites that comprise each route. Overflights of these sites will be conducted from fixed wing planes, over a four hour period, two hours on either side of mean low tide. Surveys will be conducted during the peak molt period in August. Observers will first obtain a visual count, then use a 35mm camera, with 400 ASA color slide film and an 80-200mm zoom lens, to photograph all seals hauled out. Seals are later counted from projected slide images on a white surface. Small groups of seals (10-15) may not be photographed, as the visual count is sufficient. Environmental conditions such as time, and tide height (i.e., 'covariates') will be recorded when each site is surveyed. Four or more replicate counts will be obtained to achieve sufficient precision in the trend estimate. A separate model for each combination of covariates and trend trajectories are determined, with the final trend estimate and standard error obtained as a weighted average of trend estimates from the individual models (Small et al. 1999).

**Product:** Harbor seal population trend estimates for each of the trend route areas; i.e., Ketchikan, Sitka, Prince William Sound (PWS), the Kodiak Archipelago and Bristol Bay. Trend estimates can be calculated for the entire time period for which counts are available, as well as shorter periods, e.g., the most recent 5-year period.

**Five-year project status:** Aerial survey routes with representative haulout sites for trend analyses have been established for Ketchikan (1983), Sitka (1983), the Kodiak Archipelago (1993), PWS (1984), and Bristol Bay (1998). These trend routes will be surveyed annually until the weighted standard error associated with the trend estimates is $< 1.0$, at which time a biennial survey schedule will be considered. The Ketchikan route was shifted to a biennial schedule following the 1996 survey; a survey was conducted in 1998 and the next survey is planned for 2000. Trend estimates will be calculated annually once counts are obtained.
Funded FY00.

**Project lead:** ADF&G

**4. Covariate analysis**

**Objective:** The aim of this research is to account for the influence of environmental conditions on counts obtained during aerial surveys in order to calculate more robust estimates of population abundance and trends.

**Justification:** Environmental conditions such as time of day, time relative to low tide, tide height, and date are known to influence the number of seals hauled out (Frost *et al*. 1999, Small *et al*. 1999). The influence of these conditions must therefore be accounted for in order to increase the accuracy and precision of population trend and abundance estimates.

**Methods:** Statistical models designed to estimate population trend and abundance will be developed that account for the variation in counts due to environmental covariates (e.g., time of day, time relative to low tide, tide height, and date). Models should determine the relative influence of each covariate on trend and abundance estimates, preferably by individual haulout site.

**Product:** This research will result in statistical models to adjust counts that will form the basis of more accurate and precise estimates of abundance and trends than could be obtained from using the unadjusted counts. The models of the covariate relationships from the application of this technique will help to refine our understanding of harbor seal behavior and life history.

**Five-year project status:** The first model to integrate the effect of covariates was applied to PWS trend counts, and has been published in a peer-reviewed journal. The second model, which is a modified/improved version of the first model, has been applied to all other available trend counts, and a draft manuscript should be completed in July 2000. A third model, utilizing Bayesian statistics has been applied to PWS trend counts, with a draft manuscript due in July 2000. Researchers at NMML are also applying covariate adjustments to the counts from the 1996 aerial survey for the abundance of harbor seals in the Gulf of Alaska. Funded FY00.

**Project lead:** ADF&G

**5. Haulout patterns and movement**

**Objective:** Document the haulout, movement, diving, and spatial-use patterns of Alaska harbor seals.

**Justification:** Knowledge of diving behavior and the spatial and temporal patterns between at-sea foraging areas and haulout sites will provide a better understanding of several basic life history characteristics of harbor seals, including foraging ecology, general movement and haulout use patterns. This knowledge is required to assess the possible impact of various perturbations on harbor seal populations.

**Methods:** Satellite-linked time depth-recorders (SDRs) deployed on harbor seals provide estimates of seals' locations and whether they are hauled out or at sea. These data can be summarized to provide numerous indices relative to general movements (e.g., mean distance traveled between haulouts, maximal distance between any two haulouts, mean distance to at-sea locations, size of at-sea 'foraging' areas, etc.) and haulout behavior (e.g., number of haulout sites used, haulout site fidelity, haulout timing and duration, etc.). Concurrently, information on the diving behavior (e.g., number of dives, maximal depth of any dive, proportion of time at-depth, etc.) of seals is collected continuously and summarized in 6 hour 'bins'. Over 100 adult and subadult seals were tagged with SDRs in Kodiak, PWS, and SE during 1993-1996.

**Product:** Manuscripts published in the peer-review literature that describe the general movement and haulout patterns, and diving behavior of harbor seals in Alaska. Results will be pertinent to genetic research designed to identify stock boundaries, and elucidate potential interactions with commercial fisheries.

**Five-year project status:** A draft manuscript of the general movement patterns from seals tagged in 1993-1996 should be completed by fall 2000, and draft manuscripts of haulout patterns and dive behavior by winter 2000-2001. During 1997-1999, 25 pups were tagged with SDRs on Tugidak Island and a similar number in PWS; data acquisition from these SDRs will be complete in August 2000, with analyses conducted in winter 2000-2001. Adult and subadult seals will be captured and tagged with SDRs in Bristol Bay in fall 2000. NMML has summarized findings from a study of glacial fjord haulout patterns and habitat use conducted in Tracy Arm during spring 1999. TDRs were recovered from 7 of 9 seals and an analysis of diving behavior will be conducted winter 2000/2001 in collaboration with the Canadian Department of Fisheries and Oceans. In August 2000, NMML will deploy VHF tags and a remote receiving station in Nanvak Bay (Cape Newenham, NW Bristol Bay) to measure the haulout behavior of molting seals using sandy substrate; subsequent aerial surveys will assess on a broad scale any regional movement that may occur. Funded FY00.

**Project lead:** ADF&G and NMML

**6. Glacial survey methodology**

**Objective:** Develop methodology for estimation of harbor seal population abundance at glacial haulout sites.

**Justification:** Glacial ice represents a significant haulout substrate for harbor seals in Alaska, and thus must be included in census survey efforts conducted for statewide abundance estimates. Additionally, modified or new trend routes may include glacial ice haulouts in the future. Difficulties arise in the application of conventional aerial survey techniques to glacial ice floe substrate. Seals are

conventionally photographed from fixed wing aircraft that circle over haulout locations. In terrestrial haulout areas the substrate provides a point of reference for the observers, allowing the observers to ensure that all areas of the specific haulout have been photographed, but not duplicated. In glacial ice areas the uniformity and expansive size of the substrate is problematic for observers photographing large numbers of seals. Without specific reference points it is difficult to ensure that complete coverage is obtained and duplication is prevented. For glacial ice areas with relatively few animals, the conventional aerial photography may be appropriate. However, under circumstances where large numbers of seals are spread out over large areas, determining the areas of overlap in the resulting photographs is very difficult, and hence estimates are less reliable. In these cases, the conventional methodology produces abundance estimates, which take considerable time to generate and are of unacceptably high variation (Small 1998). Alternative methods for surveying ice substrate types are needed.

**Methods:** Alternative survey techniques employing thermal imaging or conventional imaging linked to a Global Positioning System (GPS) will be attempted and evaluated. Transects will be flown over the ice and photographs taken from a "belly-mounted" camera, linked to an onboard GPS. Estimated costs for both personnel and equipment will be examined to determine the feasibility of a complete census compared to an index to abundance derived from appropriate strip- or line-transect methodology.

**Product:** A population survey technique applicable to glacial ice haulout sites.

**Five-year project status:** Glacial haulout sites in PWS were surveyed in fall 1999 using ADF&G aircraft and medium format photographic equipment linked to an onboard GPS. This technique resulted in a very large number of photographs for the larger sites (e.g., ~200 for Columbia Bay), and the resolution was less than expected. Alternative methods will be explored during winter/spring 2000, with a survey tentatively scheduled for late spring or fall 2000. Funded FY00.

**Project lead:** NMML and ADF&G

**7. Index site counts**

**Objective:** Collect land-based counts of harbor seals on the southwest and middle beach haulout sites on Tugidak Island during the pupping and molting periods to continue long-term population trend monitoring for this index site.

**Justification:** Tugidak Island, southwest of the Kodiak Archipelago, represents a unique study site for harbor seal research in Alaska. Several beaches on the island are haulout sites for some of the largest concentrations of harbor seals in Alaska. Studies of these haulout sites were initiated in the mid 1970s by the ADF&G. Since 1994, the population of harbor seals on Tugidak Island has been consistently and intensively studied. The uniqueness of this site with regard to the number of animals that haul out on the island's beaches, the excellent view of the seals afforded by the bluffs, and the long term historical

record of land-based counts make it a focus of continued investigations on population dynamics and harbor seal biology.

**Methods:** Counts of harbor seals will be conducted from 30m bluffs overlooking the haulouts on the southern and western shores of Tugidak Island. Seals hauled out on the beach will be counted using a spotting scope and binoculars. Surveys will be conducted on a daily basis within 2 hours on either side of low tide during the pupping period in May and June and the molting period in later July through early September**.** Total counts will be made as well as counts by sex and year class (pups, yearlings, subadults and adults). Yearly counts (maximal and mean estimates) will be included in a time series of counts for this area for analyses of population trend of harbor seals in the Gulf of Alaska.

**Product:** A population trend analysis, including the effect of covariates, of all counts collected through 1999 has been completed and will be presented in a draft manuscript scheduled for completion in June 2000. The manuscript will also include an evaluation of land-based counts for documenting population trend, with a comparison to trend estimates derived from aerial surveys.

**Five-year project status:** Surveys will be completed annually during the pupping period in late May through June and the molting period from late July through early September**.** Funded FY00.

**Project lead:** ADF&G

**8. Evaluation and review of survey experimental design**

**Objective:** Evaluate the experimental design used to monitor harbor seal numbers in the *Exxon Valdez* spill area and subsequently develop a revised experimental design for long term monitoring of harbor seal populations in that area, with applications for survey efforts statewide.

**Justification:** Assessment of population trend and abundance of harbor seals in Alaska is fundamental to understanding population status. Prior to the early 1980's, monitoring of harbor seal numbers occurred in only a few areas outside of Tugidak Island, and surveys were infrequent and not standardized. Beginning in 1983-84, standardized population trend surveys were established in PWS and Southeast Alaska, followed by the establishment of trend survey routes for the Kodiak Archipelago in 1993 and Bristol Bay in 1998.

Information from the trend and abundance estimates indicate population status varies across the state. Harbor seals declined by approximately 90% between 1976 and 1992 on Tugidak Island, south of Kodiak Island (Pitcher 1990, Lewis et al. 1996) and by 63% between 1984 and 1997 in PWS (Frost et al. 1998). These monitoring results have provided valuable information regarding the population status of harbor seals in the state. However, the original trend survey routes were determined primarily on logistical constraints, not on statistical sampling designs. Advances in harbor seal biology and life history as well as overall advances in marine mammal survey methodology raise new questions as to

whether the current design for population monitoring is adequately and robustly estimating trends in abundance and overall statewide abundance (Small 1999). Relatively precise and accurate estimates of population trend and abundance are required to determine appropriate management strategies. In addition, there has been no comprehensive statistical review of the population monitoring methodology for harbor seals in Alaska. A review of the survey design for the *Exxon Valdez* spill area will be applicable to other parts of the state.

**Methods:** Current monitoring programs include aerial population trend and abundance surveys, and land-based counts at a key index site (Tugidak Island). Surveys occur over a variety of areas and haulout substrate types. The current experimental design to monitor population status will be evaluated based on existing information on the spatial and temporal variation in the abundance and distribution of harbor seals, incorporating recent advances in marine mammal survey and assessment methodology. Recommendations for revisions to the experimental design will be based on the relevant statistical theory of sampling and surveying and knowledge of harbor seal biology.

**Product:** A revised survey experimental design for harbor seal population estimation (e.g. trend and abundance) in the *Exxon Valdez* spill area, Alaska.

**Five-year project status:** This project was funded by the EVOS Trustee Council and is contracted to University of Alaska Fairbanks faculty members T. Quinn and M. Adkison. Development of a new experimental design will be completed, including discussions to revise current monitoring programs, by September 30, 2000. Funded FY00.

**Project lead:** ADF&G

## B. STOCK IDENTIFICATION

### Overview

Scientists at the SWFSC have been using molecular genetic techniques to investigate population subdivision and movement patterns of harbor seals in Alaska. Variation in both mitochondrial and nuclear (microsatellite) markers is being examined to resolve population structure and estimate levels of dispersal which will provide the framework for delineating stock boundaries. The different properties of the two types of marker may also determine whether separate stocks are demographically and/or reproductively independent by distinguishing between actual (i.e., emigration) and effective (i.e., interbreeding) dispersal.

Patterns of mitochondrial DNA (mtDNA) variation in Alaska harbor seals are generally clinal in nature indicating that they do not conform to a single panmictic population. The fact that harbor seals are distributed almost continuously throughout their Alaskan range, however, has made it difficult to discern

at what scale population subdivision is acting and where sub-population boundaries lie. This has necessitated the development of new techniques for analyzing the genetic data in order to resolve population structure and identify management stocks. Two quite distinct, but complementary, approaches are currently being developed. Initial findings from the analysis of mtDNA by both approaches (Section I and II) have revealed that harbor seals in Alaska are a geographically micro-stratified species, while analyses of microsatellite markers (Section III), though still in the early stages, are already revealing structure on a broad geographic scale.

Despite the extensive number of samples and amount of genetic data that has already been collected, sample size is still small in some areas and there are (notable) gaps in our sample coverage. This has been alleviated, in part, by an initiative to establish molecular techniques to extract and analyze genetic material from alternative sample types including hair, scat, and formalin-fixed tissues (Section V). Molecular genetic tools are also being used to estimate levels of genetic diversity (Section IV) and investigate mating systems and patterns of dispersal (Section VI). Diversity indices may be informative indicators of a population's evolutionary history and current ability to deal with environmental change and disease, while the resolution of harbor seal mating systems will aid in estimating effective population size, $N_{e,}$ a parameter of relevance to estimates of rates of dispersal and the delineation of stocks.

Work needs to continue on the development of analytical methods to resolving population genetic structure in continuously distributed species. Sample collection and analysis needs to continue in order to fill in important gaps and increase sample size from key areas. Further development and optimization of laboratory protocols for the analysis of alternative sample types is required, final analyses of data sets need to be completed, and guidelines for the delineation of stocks formulated.

## 1. mtDNA variation among and within stock strata defined *a priori*

**Objective:** Define geographic strata based primarily on harbor seal distribution, abundance, and movement patterns to then be tested with available mtDNA data for evidence of genetic subdivision; (2) direct sampling to increase numbers and coverage to fill gaps in current catalog; and (3) sequence and analyze new material.

**Background:** Genetic stock division studies on harbor seals by the SWFSC were initiated in 1995, supported by the NMFS Office of Protected Resources with assistance from other sources. Over the succeeding years, as sample numbers and coverage increased, a genetic picture has emerged of a geographically micro-stratified species. The null hypotheses of a panmictic population extending across the range of the species and panmixia within current PBR boundaries were both unequivocally rejected. Now, where sufficient samples are available for analyses, significant genetic subdivision can be demonstrated between haulout areas or at least groups of a few areas. This means that demographically insignificant levels of interchange occur between them. Confidence in this observation links to the fundamental property of genetic studies: they have little statistical power to discriminate population subdivision in the presence of more than a few dispersers per generation.

The implications of geographic micro-stratification create problems both for future research and for management. Because harvest is usually unevenly distributed across the range of a species, geographic micro-stratification means that take on a given traditional hunting ground will not likely be compensated for by migration from even nearby unexploited or less-exploited areas. The potential necessity of management by small area imposes the need for finely drawn boundaries. This in turn materially increases the scale of any genetic studies in terms of sample number and coverage. In the past, harbor seal stock studies have relied largely on opportunistically collected genetic samples. In the immediate future, well-designed and specifically targeted genetic sampling will likely be required in order to draw biologically realistic boundaries in areas where, for a variety of reasons, opportunistic sampling has been rare.

**Justification:** Variation within the maternally inherited mitochondrial genome is being currently analyzed. Samples for analyses have been provided from a number of sources including subsistence harvests, tagging studies, and State of Alaska and University of Alaska tissue archives. To date, a total of 749 seals have been analyzed for sequence variation in 435bp of the mtDNA control region.

Initial analyses revealed significant genetic structure over large geographic distances (>500km). Seals from Bristol Bay, for example, are genetically differentiated from seals in Southeast Alaska. Significant differentiation has also been found over distances as small as 150km, indicating that population subdivision may occur on a much smaller scale. However, this micro-stratification was only demonstrable in areas where sample size (n = 30-40) was relatively high and well distributed among haulout sites. Clearly, the potential exists in any area for harbor seals to maintain small, isolated populations for sufficient periods so that genetic differentiation can occur. This means that caution must be applied when drafting stock boundaries based on genetics in areas of inadequate sampling. It is important to emphasize that failure to demonstrate genetic subdivision may be just as likely due to having inadequate numbers of samples as to the fact that there was none to find.

**Methods:** The SWFSC will define geographic strata based primarily on harbor seal distribution, abundance and movement patterns. Other factors that may influence or reflect movement patterns will also be considered in this process. Once strata have been chosen, the sequence data set will be analyzed with accepted genetic analytical methodologies.

Following the completion of analyses of the current mtDNA data set, the SWFSC will identify areas where sampling has been inadequate. Coordination among harbor seal investigators and management authorities will be necessary to delegate which agency or institution should have the overall responsibility (and funding) to carry out the sampling and to discuss modification of requirements due to the exigencies of collecting in remote locations. Genetic samples can be gathered from harvest, live capture operations, and by non-invasive means (e.g. hair, feces).

Tissue storage and molecular techniques will be as described in previous reports. Briefly, tissue samples will be stored in 20% DMSO and saturated salt. Total DNA will be extracted and archived using

standard protocols. Following quantification of the DNA, a section of the mtDNA genome containing the highly variable control region will be amplified by the PCR. Both strands of the target region will then be sequenced and analyzed on an automated sequencer. The resulting 435bp sequence will then be aligned and serve as the raw data for subsequent analyses of stock structure.

**Product:** A series of reports and a scientific manuscript or manuscripts detailing the genetic stock structure of Alaska harbor seals based on patterns of mtDNA variation. The analyses will be based on strata (putative stocks) defined using biological data (i.e., non-genetic data).

**Five-year project status:** A thorough analysis of the entire mtDNA data set is expected to be completed by the end of May 2000. Interpretation and write-up of the SWFSC's findings is planned for July 2000. At that time future sampling requirements will be presented to the responsible party(ies), and timelines will be refined at that point. A scientific manuscript describing the mtDNA stock structure analyses based on the current data set and biological strata determined *a priori* will be completed prior to the next SRG meeting in November 2000. Funded FY00.

**Project lead:** SWFSC

**2. *A posteriori* determination of mtDNA stock boundaries**

**Objective:** (1) Publish a manuscript describing problems with *a priori* stratification schemes applied in the absence of adequate concordant biological data; (2) develop, implement, and publish a method and software that uses genetic data to cluster most similar populations based on reasonable geographic "connectivity" rules; and (3) use such information on the mtDNA data to establish boundaries in Prince William Sound and to estimate dispersal across them by combining the genetic data with abundance estimations. The latter exploits the relationship between dispersal, population size, and genetic differentiation between strata.

**Background:** See Section 1. of Stock Identification.

**Justification:** As described in Section 1 above, analysis of stock structure has typically been done by testing the null hypothesis of panmixia (every individual has an equal chance of mating with every other individual) against the hypothesis that there is population structure (i.e. the population is not panmictic). This technique requires that the scientist begin with an *a priori* definition of strata. However, the hypothesis testing approach can easily lead to making management units too large because statistical power increases with strata size. This leads to the assertion that interpretation of statistically significant genetic results can be stated in terms of the minimum number of populations subdividing a species but using caution in the precise establishment of a particular boundary. Informal confidence in boundary placement has always come from examining concordant biological data supporting such a given scaling (i.e., data supporting limited dispersal). However, relatively continuously distributed, vagile species whose habitat has no obvious geographic barriers are particularly troublesome.

A promising approach is to use the genetic data itself to cluster the smallest supportable strata (e.g., haulout areas in the case of the harbor seal) with its most genetically similar strata.

**Methods:** The effectiveness of hypothesis-testing in determining the demographic spatial structure within a region has been evaluated by using simulated data for which the underlying spatial structure is known. From that data, the average probability level, average degree of differentiation, and power to detect differentiation as a function of the number of putative populations defined *a priori* by the researcher was calculated. Finally, the degree to which the accuracy of boundary placement corresponding to these putative population boundaries was determined. The results are that statistical power and average estimated genetic differentiation are much higher and the average probability value, much lower when a region is divided too coarsely than when it is divided in a way that accurately reflects the underlying population structure, that is known in the simulation. There is an inverse relationship between power and the number of putative populations defined. Thus, in the absence of underlying biological data informing the selection of strata, there is a danger that investigators err on the side of proposing large, more statistically reliable stock units as opposed to small, potentially more biologically realistic and risk-averse ones. It is the more coarse subdivision that produces results that have lower, type I error rates (the rate at which the null hypothesis of panmixia is falsely rejected).

Despite the difficult nature of the problem, management still requires an understanding of population structure because the presence of genetic subdivision means highly restricted dispersal. Because definition of hypothetical populations can critically influence results in a biologically risk-prone manner, the SWFSC has developed a method that searches systematically for population boundaries without forcing an initial guess at boundary location. mtDNA sequence data is used to cluster sampling sites based on genetic similarity. As above, the effectiveness of the method is tested using data generated by simulations where the true population structure is known and using different dispersal rates and different abundances of the model populations. The performance of the method is judged by making decisions about population structure by using management criteria set out by the Marine Mammal Protection Act regarding range reduction / extirpation of local populations. For most cases, our method met management objectives more than 90% of the time.

**Product:** A series of reports and scientific manuscripts dealing with the problem of *a priori* stratification occurring in genetic analyses of populations for use in management. A similar series of reports and scientific manuscripts describing the *a posteriori* approach, a potential solution to the problem. Where adequate genetic samples as well as abundance data is available, dispersal rates based on genetic data will also be estimated.

**Five-year project status:** A scientific manuscript describing problems with *a priori* stratification schemes applied to continuously distributed populations has been submitted to a scientific journal. Preliminary analyses using the *a posteriori* method for establishing boundaries between populations are underway. Actual estimates of dispersal between established boundaries will be made in areas where

there has been sufficient genetic samples collected and abundance estimates made. These estimates will perhaps be available for the November 2000 SRG meetings depending on getting workable abundance estimates sufficiently in advance to do the time-consuming simulations required. Funded FY00 (not by NMFS).

**Project lead:** SWFSC

**3. Stock identification - microsatellites**

**Objectives:** (1) Use the geographic strata developed for the mtDNA study to look for evidence of genetic subdivision; (2) direct sampling efforts to increase numbers and coverage to fill in gaps in current catalog; (3) determine the relationship between interbreeding and dispersal patterns among sub-populations; and (4) analyze new material.

**Background:** Microsatellites are a class of highly variable nuclear markers that have revolutionized the study of breeding systems, social organization and population structure. In contrast to the maternal inheritance of mtDNA haplotypes, microsatellite alleles are inherited from both the mother and father. Thus, by combining the analysis of variation at these loci with that of mtDNA, a more complete understanding of grouping, mating, and movement patterns may be achieved. Furthermore, the unusually high level of haplotypic diversity found within mtDNA in Alaska harbor seals has somewhat compromised the utility of this marker in resolving population subdivision, thus highlighting the need to look at other markers. In 1996 a project was initiated to determine the utility of microsatellites in resolving the stock structure of harbor seals in Alaska and preliminary findings were promising. To date over 250 samples from the entire Alaskan range of the species have been analyzed for variation at between 7 and 11 independent loci.

**Justification:** As with mtDNA, initial analysis has revealed structure on a broad geographic scale. Seals from Bristol Bay, for example, are genetically distinct from seals in the Gulf of Alaska. We have found that the number of samples greatly influences the reliability of estimates of genetic subdivision. Small sample size increases the variance in the test statistic thus increasing the probability of a type II error of falsely not rejecting the null hypothesis of panmixia. There is therefore a need to increase sample size from a number of areas. Boosting sample size and distribution will also enable us to investigate population structure on a micro-geographic scale and determine if stocks are demographically and/or reproductively independent by comparing findings from nuclear markers with those from mtDNA to distinguish between actual (i.e., emigration) dispersal and effective (i.e., interbreeding) dispersal. Individual microsatellite loci vary in their ability to reveal population structure, a feature that is related, in part, to how polymorphic they are. It is therefore necessary to continue to screen for variation at a large number of independent loci with differing levels of polymorphism.

**Methods:** Many samples are already available at SWFSC but new samples are required from several areas. Samples will be gathered from harvest, live capture operations, and by non-invasive means (e.g.

hair) where possible, and the collection of samples will be co-ordinated with the relevant agencies and institutions. In some areas, it will be necessary to have a directed sampling program where we will live sample seals and/or collect hair (and if nothing else is possible, faeces), preferably during the breeding or molting season when seals are most accessible and movement patterns are most restricted.

Tissue storage and molecular techniques will be as described in previous reports. Briefly, tissue samples will be stored in 20% DMSO and saturated salt. Total DNA will be extracted and archived using standard protocols. Following quantitation of DNA (and sequence analyses of mtDNA, see part 1), alleles at a minimum of 11 polymorphic microsatellite loci will be amplified by the PCR, separated on an automated sequencer, and sized with the aid of Genescan 3.1 software.

As with the mtDNA study, geographic strata to be tested with the microsatellite data will be based primarily on harbor seal distribution, abundance, and movement patterns. Other factors that may influence or reflect movement patterns are also being considered in this process. Frequency-based ($F_{st}$, $?^2$) statistics will be used to assess levels of genetic differentiation.

**Product:** A series of reports and a scientific manuscript or manuscripts detailing the population genetic structure of Alaska harbor seals based on patterns of microsatellite variation, and how this pattern compares with the mtDNA findings. The analyses will be based on the strata used in the mtDNA studies.

**Five-year project status:** The lab work on the macro-geographic structure is expected to be completed by September 2000. Interpretation and write up is planned for June 2001. Completion of lab work exploring the utility of microsatellites in uncovering micro-geographic structure within regions in Alaska is planned for March 2001. At that time decisions will be made regarding the future of this line of research and future sampling requirements will be presented to the responsible party(ies), and timelines will be refined at this point. Funded FY00

**Project lead:** SWFSC

**4. Estimates of genetic diversity as indicators of population fitness**

**Objective:** Establish whether estimates of genetic diversity at multiple loci are good indicators of population evolutionary history and fitness.

**Background:** As well as revealing population genetic structure, molecular genetic techniques can be used to investigate the consequences of population decline on spatial and temporal patterns of genetic variation. Rapid population declines can result in the loss of important genetic heterozygosity (variation) which may affect individual and population 'fitness' and compromise a population's ability to respond to environmental change.

**Justification:** The genetic consequences of dramatic declines in harbor seal abundance in certain areas of Alaska could be manifested as a reduction in reproductive capacity and lowering of the ability to deal with disease or environmental change. Caution is needed however, when using estimates of genetic diversity at neutral markers as indices of fitness, as low levels of genetic diversity may be due to natural spatial organization and mating systems instead of severe reductions in population size. Nevertheless, estimates of diversity at several independent loci may be informative when used with ecological data in determining the relative importance of environmental and genetic factors in not only causing population decline, but also inhibiting population recovery. Some preliminary comparisons have been made between Alaska populations and other harbor seal populations.

**Methods:** Once the collection of mtDNA and microsatellite data has been completed we will calculate various indices of genetic diversity. Estimates will then be compared among stocks with different abundances and trends, dispersal rates and gene flow (see parts 1, 2, and 3) in order to assess the relative contributions of evolutionary history, abundance, rate of decline, gene flow, and spatial organization and mating systems on genetic heterozygosity. We will also attempt to compare our values with those of studies on other harbor seal populations in the north Atlantic as well as north Pacific. In some cases it may be necessary to account for slight differences in the choice of markers among studies. Estimates of heterozygosity will also be compared between sample sets collected in the 1970s prior to the recent declines in abundance and sets collected in the 1990s.

**Product:** A report on the analysis of the relationship between genetic diversity at several neutral markers and recent trends and current abundance in a number of harbor seal stocks in Alaska.

**Five-year project status**: Sufficient data on 12 markers should be available by March 2001 allowing preliminary results to be available by September 2001. The analysis will be constantly updated and refined as sample size increases and sample coverage improves. Unfunded FY00.

**Project lead:** SWFSC

**5. Alternative approaches to sample acquisition**

**Objective:** Continue to develop lab techniques to extract DNA from alternative sample types in order to increase sample coverage and numbers.

**Background:** Harbor seal samples for genetic analysis have, up to now, consisted almost entirely of tissues (skin, muscle, liver) taken directly from live or dead animals. These samples come primarily from subsistence harvest and tagging operations, and as such sample coverage is dependent on where these activities take place. Because of their typically skittish nature and their tendency to haul out on relatively inaccessible coastlines, harbor seals are often difficult to catch and sample. This difficulty in directly sampling harbor seals explains, in part, the gaps in sampling along their Alaskan range and prompted us to investigate a number of indirect methods to sample acquisition.

**Justification:** Beginning in 1997 a project was initiated to develop laboratory methods to extract DNA from three alternative sources of genetic material: scat, shed hair, and formalin fixed tissues. Since then, a protocol has been developed to extract DNA from seal scat of a quality adequate enough for amplification and sequencing of mtDNA control region and analysis of variation at several microsatellite loci. Secondly, we have been successful in extracting, amplifying and sequencing mtDNA from hair shed by seals while hauled out on glacial ice in Glacier Bay. Finally, the fixing of tissues in formalin has long been accepted as excluding such samples from genetic analysis, primarily because of the difficulty in extracting DNA of high enough quality in sufficient quantity. Problems may also arise at the amplification (PCR) and sequencing stages. Considering the wealth of marine mammal samples that have been collected over several decades and fixed in formalin and subsequently preserved in alcohol, SWFSC initiated a project to develop methods to extract DNA from formalin-fixed tissues. Initial results are promising. We successfully extracted and sequenced mtDNA from 9 harbor seal samples collected from the Pribilof Islands more than 20 years ago, more than doubling our sample from this area.

These three projects have expanded greatly our ability to collect samples for genetic analysis and fill in gaps in our sampling coverage of Alaska harbor seals. Such non-invasive approaches also avoid the possible disruptive effects of traditional sampling methods on harbor seal behaviour. Further optimization of protocols is required, however. In some tissue types, sequencing is possible but microsatellite amplification is proving problematical.

**Methods:** A range of standard and novel lab techniques are being used to extract DNA of sufficient quality and quantity for PCR and sequencing from shed hair, scat and formalin-fixed tissues. Detailed descriptions of protocols will be available upon completion of this work. Briefly, methods range from standard protocols to lyse cells and digest proteins with EtOH DNA recovery to the use of guanidium thiocyanate and multiple washes to extract DNA.

**Product:** A series of theses, reports and scientific manuscripts detailing protocols will be written up upon completion. Assessments of the efficiency of each method and thus utility of each sample type will also be made.

**Five-year project status**: A complete analysis of the methods used to extract and analyze genetic material from scat and hair samples will be complete by September 2000. Progress on the formalin-fixed tissues will be reviewed in September 2000 and written up April 2001.
Partially funded FY00.

**Project lead**: SWFSC

**6. Harbor seal mating systems and dispersal patterns**

**Objectives:** Determine the mating system of Pacific harbor seals in Puget Sound and relate findings to

the analysis of stock structure of the species in Alaska.

**Background:** Little is known about the mating system of Pacific harbor seals, primarily because of the difficulty in observing mating in the wild and the limitations of using individual mating success in estimating reproductive success. Although female reproductive success can be measured directly in terms of pup production and survival, male reproductive success is impossible to determine from observation, particularly as mating frequency may not be a good index of the number of offspring an individual male fathered. With the advent of modern molecular genetic tools such as DNA fingerprinting it is now possible to accurately measure male reproductive success and thus determine the mating system of a species by estimating the variance in male reproductive success. This study of mating systems in Puget Sound may be applied to populations in Alaska.

**Justification:** Resolving the relationship between reproductive success and mating frequency can aid in the estimation of effective population size ($N_e$), an index of relevance to investigations of stock identity and dispersal in this species. $N_e$ is smaller than N, for example, if only a proportion of adult males contribute to next years cohort of pups. A small $N_e$ in turn increases the rate of genetic divergence among strata due to the greater effects of genetic drift. We are currently using a molecular genetic approach to studying the mating system of Pacific harbor seals. The combination of behavioral observations during the pupping and mating season and molecular genetic analyses of mother-pup pairs and adult males has yielded the first clear evidence of variation in male reproductive success in this subspecies. Current efforts are directed at using these findings in estimating $N_e$ for application in genetic stock identification. As well as estimating the variance in male reproductive success by establishing paternity, the same techniques are being used to assess relatedness among all members of the population. This approach will hopefully help us identify immigrants and thus describe the nature of dispersal across stock boundaries. One year of data has been collected from one location in Puget Sound to date and ideally multiple years of data at a number of different sites need to be collected to address these questions.

**Methods:** The location or locations will be chosen based on a number of criteria including the accessibility of the seals, the size of the population, and the physical characteristics of the haul-out site and surrounding area. Behavioral observations will be conducted to determine population size, record associations, and assess the mating system. Samples will be collected either directly by biopsy during capture operations or indirectly from scat, shed hair, afterbirth, or shed blood. Samples will be preserved in a 20% DMSO and salt solution or snap frozen and returned directly to the lab.

An array of laboratory procedures, many developed at our lab, will be used to extract, amplify and sequence an array of genetic markers, including mtDNA, microsatellites, and gender. (See sections 1, 3, and 5 above for details). Paternity assessment and relatedness will be estimated using a number of standard statistical packages as well as a number of techniques currently under development in our group.

**Product:** A series of theses and reports and a scientific manuscript or manuscripts detailing the findings of the current research as well as proposed research on harbor seal mating systems and how this relates to the analysis of population structure in this species in Alaska.

**Five-year project status:** A masters thesis on the current study of Pacific harbor seal mating systems is expected to be completed July 2000**.** A manuscript of this work is expected by March 2001. Timelines for continued research will be refined once agreement has been reached on study sites and duration. Unfunded FY00.

**Project lead:** SWFSC

## C. HARBOR SEAL HABITAT

### Overview

The following is an effort to integrate various external data that are relevant to harbor seal distribution and abundance. This integration may provide the basis for examining multi-factor relationships that might not otherwise be elucidated. The Geographic Information Systems (GIS) application will facilitate examination of spatial interactions that are of specific interest to management.

### 1. Characterize harbor seal habitat

**Objective:** Describe external factors associated with harbor seal distribution and abundance.

**Justification:** Data on harbor seal habitat is collected in association with other studies; however, no comprehensive set of habitat characteristics for harbor seals is available. Habitat data are useful to develop both research and management activities. Information of this nature can be used in research to assess whether trend survey sites are representative of the overall trend area and areas outside the trend route. These data are also useful for management purposes to aid in evaluation of impacts from development activity and to develop conservation measures in sensitive harbor seal sites. A GIS format of habitat maps will also allow for better assessment of complex interactions involving other species and other resource concerns.

**Methods:** Preliminarily, data will be compiled from existing sources. These data will include information on haulout substrate, bathymetry, major fishing areas, vessel traffic lanes, glacial ice areas, estimated subsistence mortality level, estimated mortality incidental to commercial fisheries, major freshwater streams, locations of human coastal communities, and an index to shoreline density (i.e. inside vs. outside waters). Specific habitat data may also be actively collected. These data will be consolidated into a GIS format and related to the spatial distribution and abundance of harbor seals in Alaska, including individual haulout and pupping locations, and the estimated home-range size and movement patterns of seals associated with particular haulouts.

**Product:** A GIS database of habitat characteristics associated with distribution patterns and abundance of harbor seals in Alaska.

**Five-year project status:** Statewide distribution and abundance data have been compiled, as have haulout substrate and bathymetry. Sources and availability of remaining habitat characteristics will be determined by early summer 2000. Integration into a GIS format will follow.
GIS formatting unfunded FY00.

**Project lead:** ADF&G

## D. HEALTH AND CONDITION

### Overview

An understanding of the health and condition of harbor seals is a basic and fundamental requirement for effective management and conservation. Research in this section is directed towards long-term studies to assess the potential impact of both natural and anthropogenic factors that may affect harbor seal health and condition. Task 1 outlines an experimental design and the methodology that can be applied towards determining current contaminant loads; whereas Task 2 describes how antibody prevalence in blood sera will continue to be analyzed for exposure to numerous disease agents. Task 3 is aimed at further developing baseline blood chemistry and hematology reference ranges to be used as general indicators of population health.

### 1. Contaminant analysis

**Objective:** Determine current contaminant loads in harbor seals in Alaska.

**Justification:** The contamination of harbor seals by persistent pesticides and heavy metals is one theory for the decline of this species in Alaska . In other species persistent organochlorine pollutants have been shown to produce toxicological effects, including reproductive dysfunction and immunosuppression. The body of knowledge available for other animal systems is much more extensive than for marine mammals. A recent review of contaminant literature for harbor seals (Papa and Becker 1998) illustrates that very little data exist on contaminant levels and their effects, particularly in Alaska waters. Papa and Becker (1998) conclude that the available data are insufficient to determine the status of contaminant loads in harbor seals throughout the species' range in Alaska. Because harbor seals are high on the food chain, the bioaccumulation of lipid soluble pesticides could produce adverse effects as has been seen in other marine and terrestrial mammals. A comprehensive baseline of contaminant loads for persistent organic pesticides and heavy metals is necessary to assess health impacts to harbor seals. A baseline study of contaminant loads will provide the basis for further scientific exploration into the effects of various chemical compounds on harbor seal health.

**Methods:** To examine current levels of metals and organochlorines (OC), 30 samples of blubber (from a standard area on the chest), kidney, and liver will be collected from male harbor seals from five regions in Alaska: SE, PWS, Kodiak, Aleutians, and Bristol Bay (six samples per region). Samples will be collected from subsistence-hunted seals through the biosampling program of the ANHSC. Samples may be collected and archived at −80 Celsius until analyzed.

Age and blubber thickness of seals, and water and lipid content of samples will also be measured to increase precision of estimates and aid interpretation of results. Examination of samples from adult males will provide more precise and maximal estimates of contaminant loads than those from adult females; loads in adult females are often lower and more variable than adult males due to dumping of organochlorines during lactation (reviewed by Addison 1989, Krahn et al. 1997). Age, which is often positively correlated to contaminant levels for male pinnipeds (reviewed by Addison 1989, Miles et al. 1992), will be determined from teeth, in cooperation with the NMML. Because OC residue concentrations are inversely related to blubber thickness (as fat is metabolized the contaminants in the remaining tissue become more concentrated; reviewed by Addison 1989), blubber thickness will be an important covariate in data analysis.

Samples will be sub-sampled following protocols developed by the National Marine Mammal Tissue Bank to prevent contamination of samples. After sub-sampling, all excess samples that are guaranteed uncontaminated will be archived at the National Marine Mammal Tissue Bank to be available for future contaminant work. Other excess tissue will be frozen at -80 at the ADF&G. Levels of 209 PCB congeners, total PCB, DDT and metabolites, and 22 other OC pesticides in blubber; and levels of 19 trace metals in kidney and/or liver will be analyzed by an appropriate analytical laboratory which participates in the quality assurance program at the National Institute of Standards and Technology.

Collection of 30 samples will reduce costs and provide current numbers for preliminary comparison with published harbor seal data from Alaska and elsewhere (Papa and Becker 1998). A power analysis has demonstrated, however, that a sample size of 200 would be needed to statistically detect geographic variation in contaminant loads (for an alpha level of 0.05, power of 0.80, and a "medium" effect size of geographic region on contaminant loads). If a preliminary comparison of metal and OC levels for 30 samples show high levels or variability of some analytes, collection of a larger sample and implementation of a statewide contaminants monitoring program will be considered.

**Product:** Baseline data set on contaminant levels in harbor seals throughout their range in Alaska.

**Five-year project status:** ADF&G will focus on the collection of samples through the biosampling program and live captures. Additionally, opportunities to work with cooperators involved with contaminant research in Alaska will be pursued to share in the cost of analyses. Initially, samples will be collected in fall/winter 2000, and 2001 (if sample size in 2000 is insufficient for certain regions). Reporting of results to the ANHSC, hunters, and interested parties will be within 6 months after all samples have been

analyzed. Unfunded FY00.

**Project lead:** ADF&G

**2. Disease/Pathology**

**Objective:** Determine baseline disease exposure in Alaska harbor seals.

**Justification:** The presence of disease in harbor seals could potentially have a significant impact on survival and reproduction, and thus population status. Few published data are currently available on disease exposure and occurrence in Alaska harbor seals. Studies indicate that harbor seals have been exposed to phocid herpesvirus, phocine distemper virus, *Brucella* spp., *Toxoplasma gondii*, and *Clamydia psittaci* (Sheffield *et al*. 1997). Although stranded animals are tested for some disease agents, a comprehensive study designed to determine the prevalence of disease in Alaska harbor seals has not been completed.

**Methods:** During 1978-1999, ADF&G obtained more than 450 sera from harbor seals collected and captured in the Bering Sea, the Kodiak Archipelago, PWS, and SE Alaska. These sera were collected to determine the antibody prevalence of selected microbial disease agents. A preliminary summary of the analyses to determine antibody prevalence was reported by Sheffield *et al*. (1997). Additional results have since been made available, and samples are currently being analyzed for phocid herpesvirus and phocine distemper virus. Once results are available from these most recent tests, ADF&G disease specialist (R. Zarnke) will complete his interpretation of all results to date and then work in cooperation with ADF&G harbor seal biologists to complete a manuscript for publication. Additional sera samples, from all possible sources, will be collected and archived for future analyses.

**Product:** A peer-reviewed manuscript published in the scientific literature that documents evidence of Alaska harbor seal exposure to eight disease agents: canine distemper virus, phocine distemper virus, phocid herpesvirus 1, *Toxoplasma gondii*, influenza A, *Brucella* spp., *Chlamydia psittaci*, and caliciviruses. Subsequent reports providing current evidence of exposure to disease agents.

**Five-year project status:** Analyses of all sera samples collected during 1978-1999 completed by March 2000, with draft manuscript completed by fall 2000. Analysis of additional sera samples collected after 1999 conducted on an as-needed basis. Funded in FY00.

**Project lead:** ADF&G

**3. Health Index**

**Objective:** Use blood parameters as a potential indicator of health status and develop statistical criteria to detect perturbations in blood parameters in populations of harbor seals.

**Justification:** Previous research on declining numbers of pinnipeds in Alaska waters elucidated the potential for detecting environmental perturbations using blood chemistry or blood proteins (Fadely et al. 1997, Zenteno-Savin et al. 1997). Most current hypotheses concerning the decline in the Alaska harbor seal population include a lower nutritional prey base. Recent studies that compared physiological and pathological parameters between stable and decreasing adult harbor seal populations in Alaska have shown some correlation between individual seal's health condition and changes in prey availability (Fadely and Castellini 1996). To determine an adult harbor seal health index using blood chemistry and morphometrics, Fadely et al. (1998) developed a method to interpret potential population "outliers" in adult harbor seals between areas of concern. To expand this concept, the health status in harbor seal pups will be examined by documenting blood parameter differences, along with morphometric measurements, in an area of chronic decline (Prince William Sound) and an area with slightly increasing numbers (Tugidak Is). It is well documented that there is a great amount of maternal transfer in harbor seals, thus using pups would provide important information on the health of pups and breeding females. Statistical methods will enable detection of developmental changes and potential "outliers" and also determination of clinical trends on the population level (Trumble et al. 1999). It must be stressed that these techniques will not be used to clinically diagnose individual harbor seals as unhealthy, but rather used to detect possible health trends in populations based on blood chemistry perturbations.

**Methods:** Develop specific baseline blood chemistry and hematology reference ranges, to be used as a health indicator, for areas of concern (i.e. declining populations) in Alaska waters. Blood from harbor seals will be drawn into various Vacutainer blood container tubes (heparinized, EDTA, and serum). Whole blood will be centrifuged and the plasma separated and frozen at -80C. Blood smears made from EDTA tubes will be used for differential counting. Approximately 1 mL heparinized plasma will be used for determination of standard plasma chemistries: sodium (Na), potassium (K), chloride (Cl), calcium (Ca), phosphate, cholesterol, glucose, protein, blood urea nitrogen (BUN), albumin, creatinine, globulin, bilirubin, lactate dehydrogenase (LDH), alanine aminotransferase (ALT), aspartate aminotransferase (AST), creatinine phosphokinase (CPK), gammaglobulin transferase (GGT), and alkaline phosphatase (AP).

Novel statistical methods will be applied to determine potential health outliers in populations of harbor seals. These models are new applications of outlier theory, principle component analysis and survivorship statistics to marine mammal management issues. They will build on the work of Fadely et al (1999) with harbor seals, Bowyer et al. (1999) with river otters and Wells et al. (in development) with dolphins. (This work is the primary focus of the Ph.D. thesis of Steve Trumble).

**Product:** Reference ranges for blood chemistry parameters in spatially distinct harbor seal populations in Alaska waters. Statistically sensitive methods for determining environmental or human based perturbations in blood chemistry and hematology values, which would subsequently be used as a potential health indicator on the population level.

**Five-year project status:** At least three years' baseline data (FY00-02) will be collected to establish meaningful blood chemistry parameters; subsequently blood parameters will be collected biannually in areas of concern. Funded in FY00.

**Project lead:** ADF&G/UAF

## E. FOOD HABITS

### Overview

Although harbor seals are known to eat a variety of fish and invertebrates (Imler and Sarber 1947, Wilke 1957, Pitcher 1980a), a complete understanding of diets throughout Alaska is lacking. Diet varies seasonally, regionally, and probably annually (Imler and Sarber 1947, Pitcher 1980a), but data on these variations are largely incomplete (Hoover-Miller 1994). The most comprehensive food habits study was conducted in the Gulf of Alaska where 548 seals were collected (269 of which had food remains in the stomach) during the 1970s (Pitcher 1980a), the largest number of samples came from the Kodiak area (n=102) and Prince William Sound (n=83). Overall in the Gulf of Alaska, the five top-ranked prey were walleye pollock, octopus, capelin, eulachon, and Pacific herring (Pitcher 1980a). The current studies listed below include complementary analyses of diet from scat and stomach contents and from fatty acid profiles extracted from blubber samples.

### 1. Food item analysis

**Objective:** Determine temporal and spatial dietary prey composition for harbor seals in Alaska, through analysis of stomach and scat contents.

**Justification:** The food limitation hypothesis (expressed as reduced availability of prey or differences in prey quality and composition) is one of several that are being investigated to elucidate ultimate cause(s) of a decline in harbor seals numbers over the last 20 years. Sporadic diet composition studies have been conducted since the mid 1940s (e.g., Imler and Sarber 1947, Wilke 1957, Pitcher 1980a). Current and comprehensive diet data, however, are lacking and for some regions, non-existent. Investigation of harbor seal diet in regions of stable or increasing populations (Southeast Alaska) and of depressed populations (Prince William Sound, the Kodiak Archipelago, and northern Bristol Bay) would allow for comparisons among areas of differing population response as well as some comparisons with historical diet data. Seasonal comparisons of primary prey are limited. Prey analysis can be accomplished through examination of scats and stomachs, which yield information on recent dietary intake.

**Methods:** Scats will be obtained from harbor seal haulout sites and stomachs will be collected from subsistence-harvested animals. Scat will be collected in Southeast Alaska, the Kodiak archipelago,

northern Bristol Bay and the north shore of the Alaska Peninsula. Scats will be collected from each of the winter, spring, late summer/fall seasons. Stomach samples were collected opportunistically from subsistence harvested animals beginning in 1995. In 1997, efforts increased to work with hunters in Southeast Alaska to obtain stomach samples. Stomach sample collection will be expanded to other areas of the state, principally the north shore of the Alaska Peninsula. Subsistence harvested animals are typically taken in the fall through the spring in most parts of the state; therefore, stomach samples will be representative of diet composition during this time period. Temporal analyses will also be conducted comparing stomach contents from recently collected samples with historical samples from the 1970's. Samples will be sent to Pacific Identifications in Victoria, B.C., Canada for prey identification.

**Product:** Manuscript(s) published in the peer-reviewed literature that describe the spatial and temporal diet of harbor seals in Alaska, and compare primary prey in the central-western Gulf of Alaska between the 1970s and the 1990s.

**Five-year project status:** Sample collections will continue, with collection efforts focusing on geographic areas and/or seasons where data are limited. Preliminary analyses of the percent occurrence of the primary prey species and of prey "categories" will be estimated by summer 2000. Final analyses will use split sample frequency of occurrence to measure the importance of different prey categories (following Olesiuk et al. 1990 and Merrick et al. 1997) and will begin in fall 2000.
Funded FY00

**Project lead:** ADF&G

**2. Fatty acid analysis**

**Objective:** Determine temporal and spatial prey composition in harbor seal diets through analysis of blubber fatty acid signatures.

**Justification:** Studies to elucidate the suite of prey species consumed by pinnipeds have focused, to a large degree, on scat and stomach content analysis. These techniques can yield useful information on recent dietary intake and on the types of prey that are consumed. However, information from these samples may be limited in that stomach and scat contents may not provide a comprehensive profile of prey consumed. Prey is digested differentially, and some diagnostic hard parts may be retained in the stomach and thus over-represented in stomach contents (Pitcher 1980b, Harvey 1989). Also, stomach and fecal material likely only reflect the contents of a recent meal from a particular area and may not represent the temporal or spatial variation of foraging efforts and prey consumption. Recent techniques of fatty acid signature analysis have been applied as an additional, and complementary, method of studying the diet composition of harbor seals (Iverson *et al*. 1997). Fatty acid signatures in prey species have been shown to be reflected in the lipid profile of higher trophic level predators. Core samples of blubber from harbor seals may yield more detailed information on diet that might not be obtained from other methods. In addition, studies in the Gulf of Alaska (Iverson and Frost 1997)

indicate that seals can be characterized to area and age class based on blubber fatty acid signatures. An understanding of shifts in diet composition over time and space and in specific cohorts may contribute to an understanding of area-specific population declines.

**Methods:** Blubber samples have been collected from harbor seals live-captured in Prince William Sound, the Kodiak region and Southeast Alaska. Additional blubber samples are obtained through the biological sampling program. Prey samples will also be collected to obtain prey fatty acid profiles for comparison to fatty acid profiles found in harbor seal blubber. Blubber samples will be processed and fatty acid signature analyses will be conducted in the laboratory of Sara Iverson, Dalhousie University, Canada.

**Product:** A manuscript published in the peer-reviewed literature describing geographic differences in the diet of seals based on fatty acid signature analyses. This work complements and enhances the work that has been conducted in Prince William Sound and it is anticipated that Sara Iverson will be lead author on such a manuscript. A manuscript or report comparing stomach content analyses and fatty acid signature analyses as methods to determine the diet of harbor seals.

**Five-year project status:** Blubber sample collection by subsistence hunters and during seal captures are ongoing. A prey collection program is underway in Southeast and is expanding to the Kodiak area to obtain prey samples for comparison with the prey library developed for PWS. Fatty acid analyses of harbor seal blubber and prey samples from Southeast and Kodiak are conducted as time allows. Funded FY00.

**Project lead:** ADF&G

## F. LIFE HISTORY/GENERAL BIOLOGY

### Overview

Information on some aspects of harbor seal life history in Alaska is limited. In particular, detailed data on the timing of pupping and molting are not available from some areas of the state. Additionally, in Southeast Alaska, the location of major terrestrial pupping sites is not well documented. Knowledge of critical habitat areas (such as large pupping sites) as well as pupping phenology is necessary to track changes in the timing and/or location of parturition and to establish a site in Southeast where disturbances can be monitored during this critical period.

Harbor seals undergo an annual cycle of regeneration and shedding, or molting, of hair. Shedding generally occurs after the pupping and mating periods in adults (Scheffer and Slipp 1944, Stutz 1967,

Ling 1972) but prior to implantation of the embryo in females (Bishop 1967, Boulva and McLaren 1979). Typically, population trends and size estimates in Alaska are based on aerial surveys conducted during the molting period when presumably the largest number of seals are hauled out. Data on both regional and sex/age differences in the timing of molting should be considered when determining optimal survey periods. The following studies will provide important information on critical pupping sites in Southeast Alaska as well as detailed information on pupping and molting phenology.

**1. Pupping and molting phenology**

**Objective:** Determine timing of pupping and molting in different regions of the state by detailed studies at representative haulout sites.

**Justification:** Harbor seal numbers declined about 90% on Tugidak Island from the mid-1970 through the early 1990s (Pitcher 1990, Small *et al*. 1998). By monitoring pupping and molting phenology on Tugidak Island we have documented important changes in the timing of these annual events between the 1970s and the 1990s. The timing of pupping was 7–18 days later in the 1970s than in the 1960s and 1990s, suggesting that females may have had difficulty obtaining adequate food resources during the period of decline (Jemison and Kelly 1997). The date of the maximal count during the molting period on Tugidak Island was 11 – 33 days later in the 1970s than during 1997 – 1999 (Jemison unpub. data). The timing of the molt also varied by sex and age class and that the peak count for each sex/age class corresponded to the early stages of active molting.

Counts of harbor seals conducted during aerial surveys are used to estimate population trends and abundance; ideally, these surveys are conducted during a peak in the molting period, when the largest number of seals are on shore. The precise timing of molting, however, is not well known throughout Alaska, although there is evidence that it does vary among regions. For example, the maximal number of seals ashore during the molting period in 1998 occurred on 2 August on Tugidak Island and on 2 September in Nanvak Bay (northern Bristol Bay) (Jemison unpub. data). Abundance surveys not conducted at a similar stage of the molt among regions may not be directly comparable. Additionally, evidence from Tugidak Island suggests that the timing of the molting period can change across years. Such a change in the timing of molting could confound comparisons of abundance estimates, and increase the variation associated with trend estimates. Trend analyses of aerial counts of seals have found that certain environmental covariates (e.g., date, time of day) significantly affect counts (Small *et al*. 1998). Incorporating these covariates in the analysis reduces the variation in the trend estimate.

Land-based studies conducted during the molting period at trend or index sites in different regions of the state will provide detailed information on the timing of the molt, identifying which sex/age classes are most abundant during surveys and helping to determine the best survey window. Regional information on the timing of pupping will document shifts during this critical time period.

**Methods:** Baseline monitoring of pupping and molting phenology of harbor seals on Tugidak Island will continue during late May – June (pupping) and late July – August (molting). When additional sites

are found in Southeast Alaska and possibly in Bristol Bay, these sites will be monitored following the protocol, described below, that was established at Tugidak Island.

Observations will be made from 30 m bluffs overlooking the haulout sites. Surveys, using a 15 - 60 spotting scope and binoculars, will be conducted daily during the pupping period and 1-2 days per week during the molting period. Seals will be categorized according to sex and age class. Sex is determined by examination of the ventral side of the animal, when exposed, and by association with a pup. Those animals for which neither approach is available will be noted as "sex unknown." Age-class will be assigned as pup, yearlings, subadults, and adults. During the molting period, each seal will additionally be categorized by molt stage (see Jemison *et al.* 1998).
If and when a new site(s) is found, the methods will follow as closely as possible to those developed at Tugidak Island, although the molting phenology data will need to be collected more frequently (3-4 times per week). At a new site, the use of time-lapse photography to extend observations across a broader time period will also be evaluated.

**Product:** Detailed information on the timing of pupping and molting in several regions of Alaska will be ascertained, with the ability to detect annual shifts in these life history events over time.

**Five-year project status:** Monitoring pupping and molting phenology on Tugidak Island will be ongoing. Establishment of a new site(s) will depend on locating an appropriate site and funding. Potential new sites will be evaluated during two 1-week cruises aboard the R.V Cobb in June and July/August 2000. Funded FY00.

**Project lead:** ADF&G and NMML

**2. Pupping colony(ies) in SE Alaska**

**Objective:** Locate large pupping colonies of harbor seals in Southeast Alaska.

**Justification:** Whereas large numbers of harbor seals are known to pup on glacial ice at several tidewater glacial fjords in Southeast Alaska, including Johns Hopkins Inlet in Glacier Bay and in Tracy and Endicott Arms, information on the location of other large pupping sites and the timing of pupping in Southeast is limited. It is important to obtain information such as the location of large (i.e, 50 or more pups) pupping sites, and where possible, document when pupping occurs.

**Methods:** Distribution data, which have largely been collected during the molting period, and local knowledge, will be examined for locations of large concentrations of harbor seals. Selected sites will be investigated either by boat or aerial survey to determine whether pups are present. Based on current information, surveys should ideally be conducted from approximately 20 May through 10 June in northern Southeast AK.

**Product:** Information on the location of large pupping sites in Southeast Alaska, and when possible, documentation on the timing of pupping at these sites.

**Five-year project status:** Ongoing during May and June of 2000-2002 depending on funding. In June 2000, the NOAA ship R.V. Cobb will be used to survey potential pupping sites in Southeast Alaska. Funded FY00.

**Project lead:** NMML and ADF&G

## G. VITAL RATES

### Overview

The vital rates of survival, maturation, reproduction, emigration, and immigration are the fundamental elements of the dynamics of a population. Estimates of the vital rates are the fundamental building blocks of demographic models and they can provide valuable diagnostic information about managed populations. These estimates are often difficult to obtain for wildlife populations, requiring long-term efforts and large samples of animals. The research described below to estimate some of the vital rates of harbor seals in Alaska attempts to satisfy the demands for large sample sizes by making efficient use of samples collected from the subsistence harvest and by developing new photographic techniques that will allow large numbers of seals to be individually identified and monitored.

### 1. Female reproductive biology

**Objective:** Estimate age-specific reproductive rates of female harbor seals including age of sexual maturation and pregnancy rates.

**Justification:** The age of maturation of mammals has been shown to depend upon conditions for growth; good conditions produce relatively larger and fatter animals that mature earlier than those faced with poor conditions early in life. Trends in the mean age of maturation for a population may signify changes in resource availability (Bengtson and Laws 1985). Age-specific pregnancy rates are closely related to fecundity, a vital rate that is fundamental to population dynamics. Therefore, it is important to monitor these parameters and to take advantage of samples available from the subsistence harvest to do so.

**Methods:** Reproductive tracts including uteri and ovaries will be collected from female harbor seals taken in the subsistence harvest. Uterine scars and counts of corpora in ovaries will be used to estimate the state of maturity and recent reproductive history of each seal.

**Product:** Estimates of age-specific reproductive rates of female harbor seals.

**Five-year project status:** Specimen material is being collected in 2000 and 2001 in cooperation with the ANHSC biosampling prgram. Funding for processing and analyses of these samples will be sought in FY2001. Unfunded FY00.

**Project lead:** ADF&G

**2. Estimating age, age of maturation, and indices of growth from teeth**

**Objective:** Assess the growth patterns and reproductive histories of harbor seals using patterns in the deposition of material in teeth.

**Justification:** Growth layers in the teeth of some phocids are deposited in variable width in relation to age (Laws 1977, Bengtson and Laws 1985) and can be used to determine the age at sexual maturity or first reproduction. Preliminary investigations of sectioned harbor seal teeth show differences in annual growth increments with a transition zone (TZ) occurring at an average age of approximately 5 years (Baker and Boveng 1997). This age is consistent with the notion that the transition zone forms at the approximate time of first reproduction or at maturity. Further investigation of growth layers may prove useful in assessing growth, condition, and reproductive histories of harbor seals.

**Methods:** Teeth will be obtained from subsistence-harvested animals and dead stranded animals. A canine and a post-canine tooth is extracted and the length and weight of each tooth is recorded. The canines provide the opportunity to measure dentine growth in a standardized manner, whereas the post-canines generally provide clearer layering in the cementum for estimation of total age and age of maturation. The teeth will be cut into thin sections and mounted on glass slides. Two independent readers will assess age by counting the growth layers of the teeth and examining for a transition zone in the widths of the cementum layers. Using a digital microscope camera and an image analysis system, the widths of the neonatal dentine, the first year dentine, and when possible, the second year dentine will be measured at standardized locations along the anterior and posterior edges of the sectioned canine teeth.

**Products:** This study will provide estimates of the age structure of the subsistence harvest and the average age at maturation of Alaska harbor seals. The estimates of age are conveyed through ADF&G to the hunters that provided the samples. The study will also provide the data for comparison of an index of juvenile growth that may indicate differences in conditions experienced by harbor seals in different places or time periods.

**Five-year project status:** Evaluation of a new thin-sectioning technique was conducted in 1998 and 1999. An initial assessment of the usefulness of dentine layer widths as indices of juvenile growth is expected to be completed by June 2000. Funded FY00.

**Project lead:** NMML

**3. Individual identification**

**Objective:** Estimate survival and reproductive rates using photo-identification.

**Justification:** Population trend estimates for harbor seals indicate a decline in harbor seal numbers in certain areas over approximately the last 20 years. These estimates provide basic information on the overall population; however, they do not provide specific information on more detailed population parameters such as survival and reproductive rates. Estimates of survival rates would allow further investigation into other factors that may directly affect survival, which ultimately affect population trend. Survival estimates and reproductive rates can be obtained through mark-recapture studies, whereby an animal is uniquely identified either by being "tagged" with an external marking or by identifying a unique mark that will enable the observer to resight the animal at future time periods. Unique identifiers have been used on humpback whales by photographing the distinct pattern of notches, scars and pigmentation on the undersides of these animals' flukes (Mizroch et al. 1990). Unique identification of pinnipeds by some intrinsic marker is more problematic. However, a technique has been developed by Lex Hiby of Conservation Research Ltd. (CRL) whereby a photograph of the unique pelage pattern of gray seals, *Halichoerus grypus*, is taken, digitized and catalogued for comparison to other images in the database (Hiby and Lovell 1990). The ADF&G has begun preliminary investigation of this technique for harbor seals hauled out on the beaches of Tugidak Island. Images of over 1,500 seals have been successfully digitized and catalogued. If after testing and determining pelage patterns of individuals are consistent from weaning into adulthood, data from seals first photographed as pups will be used to estimate age-specific survival and reproductive rates. Future studies will focus on the continued collection of images from Tugidak Island and a more extensive mark-recapture study to determine survival rates will be developed.

**Methods:** Seals will be photographed on Tugidak Island using a 35mm camera and a Schmidt-Cassegrain telescope with an eight inch aperture. Additionally, high resolution video equipment will be used in an attempt to increase image quality and sample size. The sides and front of the head and the ventrum will be photographed, and images will be sent to CRL, where they will be digitized and scanned into a computer database. The best images taken for each individual seal will then be fit to a 3-dimensional model (Hiby and Lovell 1990) to correct for posture and viewpoint. The pelage pattern will be extracted and described numerically to determine that particular seal's marking pattern. Each 'mark' (i.e. seal) will then be entered into an ACCESS database, to be used for subsequent matching analyses of other 'marks'. Seals will be photographed during pupping and post-molt, data will be pooled into two sampling occasions per year (pupping and post-molt), and annual and between-season survival will be analyzed using mark-recapture models.

**Product:** Estimates of annual survival will be available within 5 years of project initiation. Estimates of population size, reproductive rate, pup mortality, and movement rates may be available each year from data collected in summer and fall. A comprehensive analysis of population dynamics of

harbor seals at Tugidak Island will be possible after long-term data (10+ years) are available. An evaluation of age-related changes in pelage patterns of harbor seals, within 5 years of project initiation, to determine if age-specific survival and reproductive rates can be estimated using photo-id data.

**Five-year project status:** Seals will be photographed annually during pupping and post-molt. Analysis of data from 1998 and 1999 has shown the 3-dimensional model of the head and matching software performs well with only a 2-3% probability of missing a match. However, a ventrum view will provide a larger sample and better demographic data because all seals photographed can be sexed. A 3-D model for the ventrum will be created and tested in winter/early spring 2000, for possible use during pupping season 2000. If the ventrum model performs well, images of the ventrum and head will be taken in 2000 to provide a transition from the head view to a ventrum view and therefore preserve data acquired in 1998 and 1999. The use of digital video will also be explored during pupping 2000. Images will be analyzed in late fall annually by CRL and survival analysis will be conducted after 4 to 5 years of data collection. Funded FY00.

**Project lead:** ADF&G

## H. HUMAN INTERACTIONS

### Overview

The research categories addressed above focus on harbor seal biology and ecology. The following research tasks examine direct anthropogenic impacts to harbor seals. Tasks 2 and 3 reflect the specific need, as mandated in the MMPA, to obtain estimates of human-caused mortality and injury to harbor seals. The two sources of direct human-induced harbor seal mortality and injury in Alaska are subsistence removals and, potentially, commercial fisheries interactions. The first task reflects the growing concern about the potential impacts to harbor seals from vessel disturbance.

### 1. Disturbance at haulouts

**Objective:** Determine the effect of vessel presence near harbor seal haulout areas.

**Justification:** Harbor seals haulout daily on a variety of substrates, including rocky islets and glacial ice. During the pupping and molting periods, seals spend more time on haulouts. Harbor seals hauled out on land or ice are vulnerable to disturbance by vessel, aircraft and land-based viewing. When hauled-out seals are frightened by approaching vessels, aircraft or people on land, they will retreat into the water. Disturbance from a resting or pupping site can increase the energetic requirements of the animal and could also result in separation of mothers and pups, breaking the mother/pup bond and reducing the chance of pup survival. Disturbance at a particular site may cause shifts in harbor seal haulout patterns and distributions (Allen *et al.* 1984).

Many of the harbor seal haulout sites, both terrestrial and ice-based, are easily accessible by wildlife viewing tours. Vessel-based wildlife viewing in coastal Alaska has increased substantially in the last 10 years. Many of the vessels (ranging from large cruise ships to small eco-tourism vessels, including kayaks) approach haulout areas to provide their passengers with the opportunity to view harbor seals in the wild and at close range. There are few studies in Alaska that have examined the effect of vessel presence and activity on harbor seal haulout behavior; no general baseline information exists to monitor changes in viewing pressure. A study on the effect of vessel disturbance is necessary to understand whether and to what degree vessel disturbance could be affecting harbor seals.

**Methods:** Observation sites will be established in several locations that experience vessel traffic. These sites may include terrestrial haulouts and ice haulouts at tidewater glacial fjords. Methods and protocol will be in cooperation with and based on pilot studies conducted by Beth Mathews through Glacier Bay National Park and University of Alaska Southeast (Mathews 1997, Mathews 1999) . Data on movements off of the haulout and behavioral changes in the animals will be collected during two periods: pupping in late May/June and molting in late July/early September. This information will be collected during times when vessels are present and when vessels are absent. Another site will be established where controlled vessel approaches will be conducted to determine the effect at different distances from the haulout and at different vessel speeds.

**Product:** Information on the effect of vessel presence and approaches on the haulout patterns, movements, and behavior of harbor seals.

**Five-year project status:** No work in progress. Unfunded FY00.

**Project lead:** To be determined based on funding source.

**2. Harvest monitoring and mortality estimation**

**Objective:** Determine the total number, including the proportion of struck and lost animals, as well as sex and age, of harbor seals harvested by Alaska Natives.

**Justification:** The MMPA requires an estimate of the annual human-caused mortality and serious injury of marine mammal stocks by source. The subsistence harvest of harbor seals represents one source of human-induced mortality or serious injury. Harbor seals are a traditional subsistence food of Alaska Natives in many coastal Alaska communities. In addition to being a food source, harbor seals represent a significant part of the cultural and spiritual basis of Native communities. Alaska Natives may take marine mammals for subsistence use under both the MMPA (Section 101(b)) and the Endangered Species Act (Section 10(e)). Native takes for subsistence or handicraft purposes are generally not subject to regulatory control unless a stock is depleted (MMPA) or unless Native takes are substantially disadvantaging the stock (ESA). Although Native subsistence harvest of harbor seals is not the subject of direct management action, monitoring of all mortality, including subsistence

removals, is necessary to ensure that the harbor seal population(s) does not fall below an optimum sustainable population.

**Methods:** Subsistence harvest levels will be estimated using either direct hunter reporting or retrospective survey techniques. The methods to be used will be determined and implemented in close coordination with the ANHSC.

**Product:** A time-series of the total subsistence takes of harbor seals in Alaska including the number of animals taken and struck and lost, by sex, age class and geographic region.

**Five-year project status:** A seven year time series of subsistence harvest data were collected by the ADF&G Subsistence Division for harvest years 1992-98. Continued harvest monitoring will be done in conjunction with ANHSC. A monitoring schedule has yet to be determined and is contingent upon securing funding. At present, long-term funding has not been secured. Unfunded FY00.

**Project lead:** Alaska Region, in consultation with ANHSC

**3. Incidental take by commercial fisheries**

**Objective:** Determine the level of incidental take of harbor seals in commercial fisheries off the coast of Alaska.

**Justification:** The Marine Mammal Protection Act (MMPA) requires that a species or population stock not be permitted to diminish below its optimum sustainable population and that measures be immediately taken to replenish any species or population stock which has already diminished below that point. Section 118 of the MMPA specifically mandates that the incidental mortality or serious injury of marine mammals, including harbor seals, occurring in the course of commercial fishing operations be reduced to insignificant levels approaching a zero mortality and serious injury rate. Fisheries are classified according to the degree of interaction with marine mammals. Should the level of human-induced mortality exceed the potential biological removal (PBR) level and the stock be declared "strategic" the commercial fisheries which interact with that species would be required to reduce the incidental mortality and serious injury of that stock taken incidentally in the course of commercial fishing operations to a level below the PBR calculated for that stock. As a result data must be collected on the level of incidental serious injury and mortality occurring in commercial fisheries. This information is also required in the annual stock assessment reports. Currently few data are available on the incidental take of harbor seals in commercial fisheries.

**Methods:** Current methods include reporting via a fisher self report system and directed observer coverage in some commercial fisheries. Federal groundfish fisheries have varying observer coverage depending on vessel size and fishing targets; selected state fisheries are observed on a periodic basis.

Estimated observer coverage levels in state fisheries are calculated based on a statistical model that incorporates fishing effort and PBR levels of a reference species.

**Product:** An estimate of the number of harbor seals taken incidental to commercial fisheries in Alaska.

**Five-year project status:** The fisher self report system and groundfish observer program continue on an annual basis. The marine mammal observer program is currently focusing on Category II fisheries (those fisheries having occasional incidental mortality and serious injury of marine mammals) in Alaska on a rotating schedule. The first year of implementation (1999) for the dedicated marine mammal observer program, observers were placed in the set and drift gillnet salmon fisheries in Cook Inlet. These areas and fisheries will be observed again in 2000. In 2001 and 2002 observer coverage will be moved to the set gillnet fisheries in the Kodiak and Yakutat areas. All incidental takes of marine mammals, including harbor seals, will be documented. Pending funding beyond 2002, fisheries in other areas of the state, including Southeast Alaska and Bristol Bay, will be observed. Funded FY00.

**Project lead:** Alaska Region

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Appendix 1

**BUDGETS**

## ADF&G

| | **AMOUNT** |
|---|---|
| **Labor** | $289,306 |
| **Operations** | $337,562 |
| **Maintenance** | $44,132 |
| **TOTAL** | **$671,000** |
| **Unfunded projects** | $20,000 |

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## SWFSC

| | |
|---|---|
| **Labor** | $128,000 |
| **Operations** | $22,000 |
| **Infrastructure** | $10,000 |
| **TOTAL** | **$160,000** |
| **Unfunded projects** | $52,000 |

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**NMML**

| | |
|---|---|
| **Labor** | $221,700 |

Appendix 1

| | |
|---|---|
| **Operations** | $200,100 |
| **TOTAL** | **$441,800** |
| **Unfunded projects** | $125,000 |

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| | |
|---|---|
| **AKR** | |
| **Labor** | $100,000 |
| **Unfunded projects** | $250,000 |
| **Other** | |
| **Unfunded projects** | $60,000 |